

## **Lung Mechanics in Marine Mammals**

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### **LONG-TERM GOALS**

The long term goal of this study is to develop methods to study lung physiology in live marine mammals and to use these techniques to investigate the mechanical properties of the respiratory system in different marine mammals. This effort is vital to understand how diving mammals manage inert and metabolic gases during diving and will help determine what behavioral and physiological responses increase DCS risk.

### **OBJECTIVES**

Recent theoretical studies have suggested that marine mammals commonly live with elevated blood and tissue N<sub>2</sub> levels, and that they use both physiological and behavioral means to avoid DCS [1, 2]. But what physiological variables are the most important to reduce N<sub>2</sub> levels below those that cause DCS, and how important is a link between behavior and physiology? For example, if the duration of each individual dive was extended, the repeated dives during a bout (a series of repeated dives with a short intervening surface interval) may result in accumulation N<sub>2</sub> to levels that may cause DCS. A variety of situations, such as sonar exposure, reduction in prey abundance or environmental change, may result in behavioral changes in dive pattern. Such changes could cause elevated tissue and blood N<sub>2</sub> levels that either result in DCS or force the animal to end a foraging bout prematurely to prevent the formation of inert gas bubbles. Prematurely ending a diving bout reduces foraging efficiency and could have detrimental implications for survival. While the results from theoretical studies have to be viewed with caution, sensitivity analyses have indicated that the degree of gas exchange and cardiac output

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during diving are the most important variables determining  $N_2$  levels in blood and tissue, and thereby the DCS risk. However, our current knowledge of how gas exchange is altered by compression of the respiratory system, possibly to the limit of collapse, is rudimentary at best.

The proposed lung collapse depth in the Weddell seal is around 30 m [3] and 70 m for the bottlenose dolphin [4]. In another study, no apparent differences in pulmonary shunt were observed between species with widely different respiratory structure [California sea lion vs. harbor seal, 5]. The results also suggested that complete cessation of gas exchange may not occur until a depth  $> 150$  m [5], even when the animal exhaled before diving. In a previous ONR funded effort, a mathematical model was developed to explain these divergent results (ONR award number N00014-07-1-1098). The results from this work implied that beaked whales commonly experience end-dive  $N_2$  levels that would cause a significant proportion of DCS cases in terrestrial mammals [1]. It was proposed that as the  $N_2$  levels increased they could eventually limit the extent of a dive bout [1, 2, 6, 7]. It was also suggested that the normal dive behavior and physiological adjustments could be important to reduce end-dive  $P_{N_2}$  [1, 6, 7].

The model results predict that the alveolar-collapse-depth, and thereby the degree of gas exchange, is greatly affected by the compliance values of the different parts of the respiratory system [8]. While results from mathematical models should be tested with empirical data, few studies have examined respiratory mechanics of live marine mammals [9, 10]. The model therefore used compliance values from an excised marine mammal lung for the lower respiratory tract, and of an excised trachea from a terrestrial animal for the upper airways [8]. To enhance our ability to predict how anthropogenic sound may interact with gas management during diving, an improved understanding of the physical properties that affect compression of the respiratory system and gas exchange is warranted.

## APPROACH

This project is separated into three aims:

**Aim 1:** We will measure the inspiration ( $\dot{V}_{insp}$ ) and expiration flow rates ( $\dot{V}_{exp}$ ) during quiet breathing in addition to airway ( $P_{air}$ ) and esophageal ( $P_{eso}$ ) pressures. These data will be used to calculate airway resistance, pulmonary and thoracic compliance (pressure-volume relationship). The static compliance values will be compared to the data previously determined in post-mortem marine mammals (Fahlman and Moore, unpublished observation). We hypothesize that deep divers have a more compliant respiratory system that will enhance compression and collapse of the thoracic cavity.

**Aim 2:** We will monitor end-tidal  $O_2$  and  $CO_2$  in anesthetized, spontaneously breathing marine mammals. We hypothesize that species that dive deeper and for longer duration have significantly lower end-tidal  $O_2$  and higher  $CO_2$  levels.

**Aim 3:** The experimental results will be compared with data obtained from our previous and on-going hyperbaric CT studies. The combined results will be used to revise a model that predicts the extent of gas exchange for a range of species.

## WORK COMPLETED

### *Aim 1:*

In the first year, we planned the experimental design and tested the procedure and research equipment. We conducted several preliminary tests (Table 1). In some animals where euthanasia was planned, we managed to measure both lung mechanics in vivo during spontaneous breathing (dynamic) and mechanical ventilation (static), and the static compliance of the excised lung after euthanasia.

*Table 1. Number of samples in each category*

Species	Live animal	Excised lung
California Sea Lion	11	6
Elephant seal	1	-

We had problems with the connections between the pneumotach and the intubation tube and we will make custom-made fittings for next year which will allow us to use 1 setup for all sizes of animals. Additional measurements will be made next year to increase the sample size and to complete aims 2 and 3.

*Aim 2:* The metabolic system that was purchased to monitor end-tidal O<sub>2</sub> and CO<sub>2</sub> was tested and did not meet specifications and had to be returned. A new system was purchased but the company could not deliver the system in time for the data collection in Sausalito.

The new system will be tested during the fall 2012 and measurements made of end-tidal O<sub>2</sub> and CO<sub>2</sub> be made in the second year.

*Aim 3:* Analysis of the data collected in the first year is underway. As we have collected respiratory compliance data in both live animals and then in the excised lungs after the animal was euthanized, we plan to compare how well these data compare. We will then compare these data to our previous data from excised lungs collected at Woods Hole Oceanographic Institution.

## RESULTS

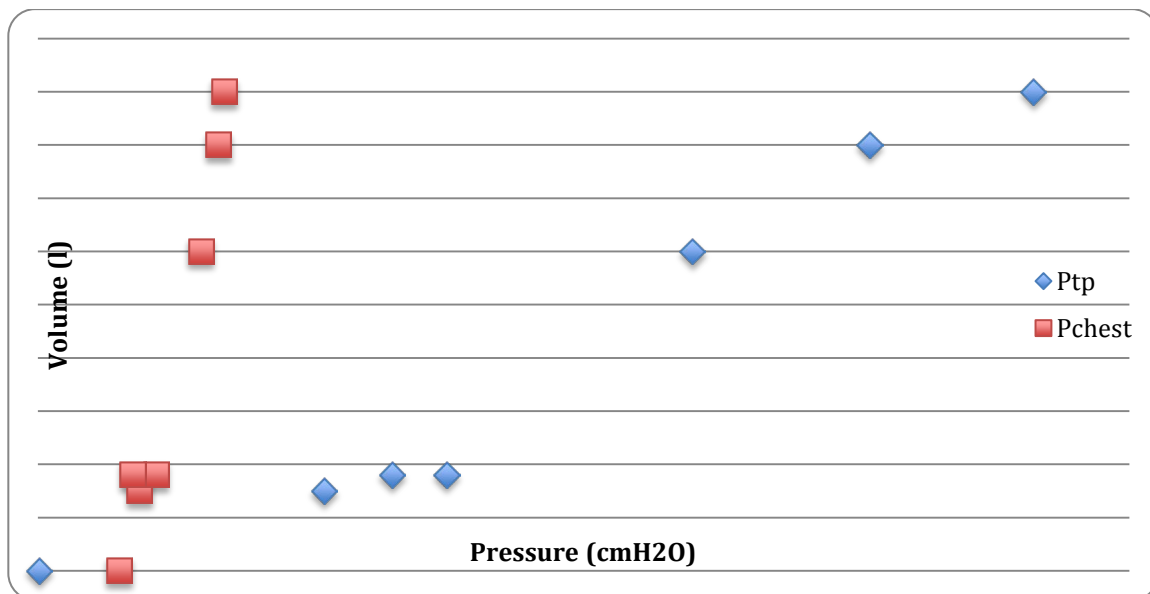
*Aim 1:* We successfully collected respiratory data from 12 animals, 1 elephant seal and 11 California sea lions. In some animals, we were able to collect data from the live animal and then from the excised lung after the animal had been euthanized. Figure 1 shows a representative sample of raw data collected from a sea lion. Figure 2 shows analyzed data for the relationship between pressure and volume during mechanical breathing in an anesthetized sea lion. The transpulmonary pressure was expressed as the difference between the airway and the esophageal pressure while the transthoracic pressure was the differential pressure between the esophageal and ambient pressure. These data indicate that the chest is much more compliant than the lung and agrees with the suggestion that the marine mammal chest provides little resistance during respiration.

## Measured lung compliance in sea lions



nimal 0.365 l x cmH2O a value half of that in a pilot whale

*[Figure 1. Raw data from mechanical ventilation of a sea lion.]*



*[Figure 2. Raw data from mechanical ventilation of a sea lion.]*

**Aim 2:** We were not able to collect any continuous end-tidal O<sub>2</sub> and CO<sub>2</sub> data due to complications with the system that was purchased. However, we used a capnograph to collect some end-tidal data in 2 sea lions. These data suggested that sea lions may experience relatively (relative to terrestrial animals) high end-tidal PCO<sub>2</sub> levels without apparent problems. In addition, we collected continuous heart rate data in one sea lion during a 3 hour cataract surgery. These data will later be used to analyze the ECG trace for heart rate variability.

**Aim 3:** The data analysis is underway (see figure 1 and 2) and we will combine these data with our previous data set in excised lungs from odontocetes and phocids. We will add the second year of measurements to the analysis at the end of the study and update our model that predicts the air volumes in the upper (conducting airways) and lower (alveolar space) airways.

## IMPACT/APPLICATIONS

This work is intended to enhance our understanding of how the respiratory system responds during diving in marine mammals. The results will provide information that will allow us to provide species specific pressure-volume parameters for the airways. These data will enable more realistic predictions of how the lungs compress to the limit of collapse and improve our understanding how marine mammals manage gases during diving.

The results can be used to determine how changes in dive behavior, including those from man-made interference, affect blood and tissue  $P_{N_2}$  levels. Thus, our results will enhance the fundamental understanding and interpretation of avoidance of the effect of anthropogenic sound, and enable knowledgeable decisions about sonar deployment, related training exercises and responses to NGO concerns. This should be of value to the US Navy Marine Mammal Program.

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